CellFree Sciences

Technology Application Note:

Vaccine Production

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Vaccine Production

One of the most promising applications of ENDEXT® Technology is to the identification of novel malaria vaccine candidates. Numerous attempt to produce effective vaccines against malaria have failed so far because of difficulties in the recombinant expression of malaria proteins.

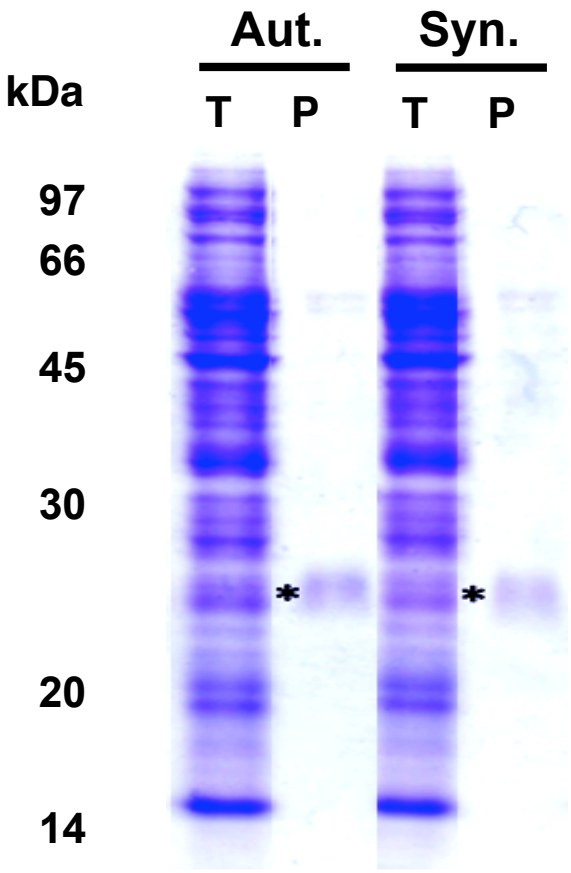
Difficulties in recombinant malaria protein expression

1. The (A+T) content in the coding sequence is as high as 76 %.
2. Correct folding is required.
3. Vaccine candidate proteins have to be found among those which are not glycosylated, because the malaria parasite proteins are not.

ENDEXT™ Technology offers genome-wide synthesis of malaria protein

1. An AT-rich malaria vaccine candidate have been successfully expressed with a proper folding by ENDEXT® Technology.
2. Antiserum against the recombinant protein contains highly protective anti-parasite antibodies.
3. ENDEXT® Technology is a powerful tool for high-throughput genome-wide screening of novel malaria vaccine candidates.

Successful expression of recombinant malaria protein using ENDEXT® Technology



Aut. :
expressed from
authentic gene (70 % AT)

Syn. :
expressed from
codon optimized synthetic gene (58 % AT)

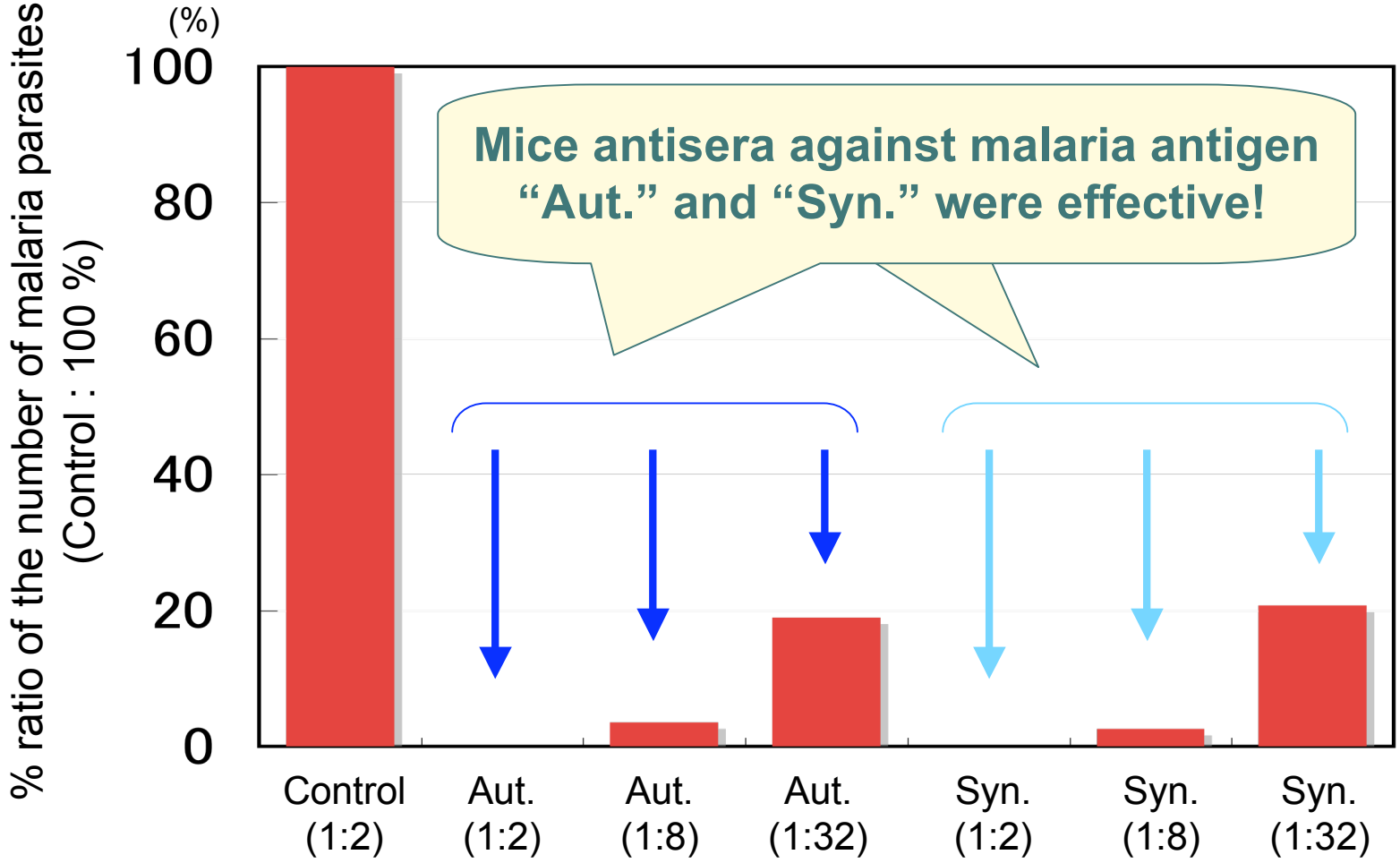
T: Total
P: Purified (Ni-NTA)

→ **Antibody production**

This figure is by courtesy of Prof. Tsuboi, Ehime Univ.

Both proteins are successfully expressed in soluble fraction by wheat germ cell-free system.

Blocking effect against malaria parasite



This figure is by courtesy of Prof. Tsuboi, Ehime Univ.

This strongly suggests that antigenic components produced by ENDEXT® Technology qualify as vaccine candidate.

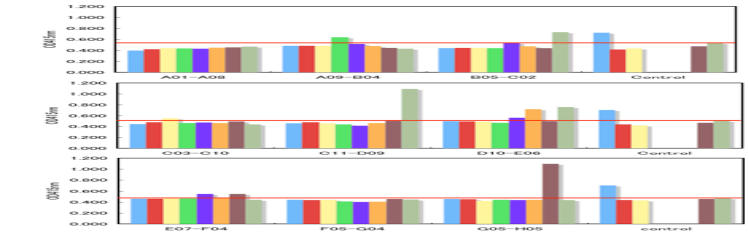
Genome-wide synthesis of malarial protein

	Genes	RT-PCR	Template	Protein Produced
WEPRO™	666	661/663	544/631	304/383
E.coli *	292			39/292

*Aguiar, et al. Genome Research 14:2076–2082, 2004

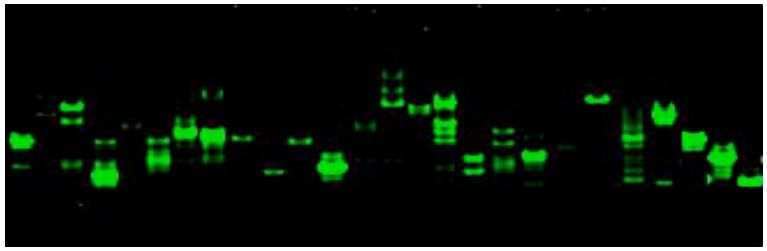
Screening of candidates using sera from the patients

ELISA



Positive

FCS



Prep. of Antigen → Antibody → Bio Assay

This figure is by courtesy of Prof. Tsuboi, Ehime Univ.

Genome wide synthesis of malarial protein

By *in silico* screening, 666 malaria genes were selected, whose template DNAs were prepared by RT-PCR.

Nearly 400 target genes were subjected to expression by ENDEXT® Technology. About 80% of the tested genes were expressed and more than half of them were mainly detected in soluble fraction, and others were both in soluble and insoluble fractions.

This result suggests that ENDEXT® Technology is a powerful tool for high-throughput genome-wide screening of novel malaria vaccine candidates.

The methodology of ENDEXT® Technology, which was successfully used for the high-throughput genome-wide synthesis of malaria protein, is also applicable to other infectious diseases such as bird flu, SARS, and West-Nile.

CFS offer contract R&D and/or collaborative R&D services for developing new diagnostic reagents and/or drugs for infectious diseases.